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Title: Vapor-Phase Decontamination of Apples Inoculated with *Escherichia coli*

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Vapor-phase Decontamination of Apples Inoculated with *Escherichia coli*

G.M. SAPERS, P.N. WALKER, J.E. SITES, B.A. ANNOUS, AND D.R. EBLEN

ABSTRACT: Improved methods of decontaminating apples containing human pathogens are required. In this study, application of gaseous antimicrobial agents was investigated. An apparatus, which transfers vapor from hot antimicrobial solutions to a treatment vessel, was evaluated with Golden Delicious apples inoculated with *E. coli*. Vapor from glacial acetic acid at 60 °C provided population reductions exceeding 3.5 log₁₀ CFU/g but induced discoloration. Vapor from heated hydrogen peroxide and chlorine dioxide solutions gave reductions ≤ 2 logs. However, chlorine dioxide gas, applied at 20 °C in an airtight container, achieved a 4.5 log reduction, with minimal quality loss. Reductions achieved with this treatment greatly exceed those obtained by other decontamination methods for raw commodities.

Keywords: apples, *E. coli*, decontamination, vapor-phase, acetic acid, hydrogen peroxide, chlorine dioxide

Introduction

PERIODIC OUTBREAKS OF FOODBORNE illness associated with unpasteurized apple cider have prompted the U.S. Food and Drug Administration to promulgate regulations for cider producers that require instituting HACCP programs and application of treatments to the juice that result in a 5-log reduction in the population of the most resistant human pathogen present (FDA 2001). A recent recall of commercial fresh-cut apples contaminated with *Listeria monocytogenes* also raises concern about the microbiological safety of fresh-cut apple products (Anonymous 2001). Conventional means of decontaminating apples and other fruits and vegetables by washing with water, detergents, or sanitizing agents are only capable of reducing attached microbial populations by 1 or 2 logs at best (Sapers and others 2000; Annous and others 2000; Wright and others 2000; Wisniewsky and others 2000). Washing efficacy may be limited by the inaccessibility of bacteria attached in the stem and calyx areas of apples or in punctures (Sapers and others 2000). Innovative decontamination treatments that provide better contact with bacteria located in inaccessible attachment sites are needed to achieve greater treatment efficacy.

Antimicrobial agents applied in the vapor phase might be capable of contacting and inactivating such inaccessible bacteria. Studies carried out at the Pacific Agri-Food Research Center (PARC), Agriculture and Agri-Food Canada in Summerland, BC demonstrated that acetic acid vapor treatment of cabbage, mung bean seeds, and grapes reduced microbial populations and prevented decay (Sholberg and Gaunce 1995,

1996; Sholberg and others 1996, 1998; Delaquis and others 1997, 1999). In a preliminary study carried out at PARC, apples were artificially contaminated with a nonpathogenic *E. coli* strain and then exposed to acetic acid vapor at 50 °C. Surviving *E. coli* populations on the apple skin and in the calyx and stem areas were determined. Exposure to acetic acid vapor for 30 min resulted in a 5-log reduction in the calyx and skin portions and a 3.5-log reduction in the stem area. However, the treated apples had an acetic acid odor and exhibited darkened and sunken lenticels and severe browning of the calyx area, indicative of injury, following refrigerated storage for 72 h (Delaquis and others 2000).

Hydrogen peroxide (H₂O₂) vapor treatments have been used to inhibit post-harvest spoilage of grapes (Forney and others 1991), melons (Aharoni and others 1994), mushrooms (Sapers and others 1995), and other commodities, and to decontaminate prunes (Simmons and others 1997). However, vapor treatments tend to be slow and can cause discoloration in some commodities such as mushrooms, raspberries, and strawberries (Sapers and Simmons 1998). These limitations might be overcome if the treatment could be applied more efficiently.

Vapor-phase decontamination of produce with chlorine dioxide (ClO₂) has been investigated (Han and others 2000, 2001a, b). Such treatments were highly effective in reducing populations of *E. coli* O157:H7 on inoculated green pepper. Treatment of potatoes with air purged through chlorine dioxide solutions reduced native microbial populations and spoilage of potatoes inoculated with *Erwinia carotovora* (Tsai and

others 2001). However 2-h treatment times were required to obtain a 2-log reduction in total aerobic bacteria and yeast and mold populations.

In the present study, our objectives were to compare the performance of a new apparatus for applying vapor-phase treatments to apples by vacuum/pressure cycling with that of a novel system for applying chlorine dioxide vapor to apples, and to determine the efficacy of such treatments in reducing *E. coli* populations on inoculated fruit without inducing injury in the product.

Materials and Methods

Source and inoculation of apples

Unwaxed Washington State Golden Delicious apples, obtained from a single grower, were stored at 4 °C until needed. In addition, Granny Smith, Gala, and Fuji apples were obtained from a produce distributor for comparison with Golden Delicious. Apples were inoculated with a nonpathogenic *E. coli* (ATCC 25922), used as a surrogate for *E. coli* O157:H7 (Sapers and others 2002). Inocula were prepared by growing the organism in 10 mL TSB at 37 °C for 8 h, transferring 100 mL of this late exponential-phase culture to 1 L of TSB, and allowing the culture to grow with shaking at 75 rpm for approximately 18 h at 37 °C. The turbid culture was then centrifuged at 3874 × g for 10 min, washed once with 200 mL sterile distilled water, and the resulting cell pellet resuspended in 2 L of sterile distilled water for a final cell concentration of approximately 8.8 log₁₀ CFU/mL (range of 8.7 to 9.1 log₁₀ CFU/mL). Apples, taken from refrigerated storage, were immediately immersed in the stationary phase *E. coli* cell

suspension at ambient temperature for 5 min, drained in a colander, and placed on their sides in plastic tubs lined with absorbent paper. The inoculated apples were held for 24 h at 4 °C prior to use to provide sufficient time for bacterial attachment and more closely simulate contamination prior to or during harvest.

Design and operation of vapor treatment apparatus

The vapor treatment apparatus described herein was derived from a system developed and used at PARC in preliminary experiments conducted there (Delaquis and others 2000). Our vacuum-pressure cycle apparatus essentially consisted of a 6-L stainless steel treatment vessel, 4 valves, and 2 horizontal 316 stainless steel pipes, 10 cm dia and 152 cm long. One of the pipes was the vapor generator, to which was added 300 mL glacial acetic acid, 1000 mL 35% H₂O₂ solution, or 735 mL of a 1200 mg/L ClO₂ solution prepared by immersion of a sachet containing proprietary precursor materials supplied by a cooperator (ICA TriNova, LLC, Forest Park, Ga., U.S.A.). The other pipe was the compression cylinder, which contained a free piston. A simplified form of the system is illustrated in Figure 1. For the acetic acid vapor trials, the entire apparatus except the top portion of the treatment vessel was submerged in a 1000-L capacity dump tank, which functioned as a water bath. The purpose of the bath was to control the temperature for the experiments and to minimize condensation of the treatment vapor on apparatus surfaces. For H₂O₂ and ClO₂ trials conducted subsequently, the entire apparatus was lowered so that the treatment vessel was completely submerged. This change was implemented to further reduce condensation, which was suspected as being a major source of variability with acetic acid vapor treatments. Apples were placed in the treatment vessel, which in later experiments contained a hemispherical

plastic colander to prevent the apples from touching the normally hot sides of the vessel. The operating cycle was begun with the piston at the far-right end of the pipe and all valves closed. Opening the vacuum system valve and V3 moved the piston to the left and created a 510 mm Hg vacuum in the treatment vessel, at which time V3 was closed. Valves V1 and then V2 were opened, flushing the vapor from the vapor generation pipe into both the treatment vessel and the right end of the piston pipe and leaving atmospheric pressure in the treatment vessel. V1 and V2 were then closed. Air pressure from opening V4 moved the piston to the right, pressurizing the vapor in the vessel to desired level. Pressure was held for a specified dwell time and then released, ending the cycle. The cycle was repeated as specified and then the apples were removed.

Application of vapor-phase antimicrobial treatments

Replicate sets of 4 inoculated apples were removed from 4 °C storage; immediately placed in the treatment vessel and exposed to the antimicrobial vapor applied using different vacuum/pressure cycles (vacuum of 508 to 686 mm Hg; pressure of 34.5, 68.9, or 103.4 kPa), numbers of cycles per treatment (1 to 4), treatment times (5 to 30 min), and temperatures (40 to 60 °C). Treated apples were immediately rinsed with tap water for 30 s, observed for treatment-induced injury, and then prepared for microbiological evaluation and for the estimation of acetic acid or hydrogen peroxide uptake (uninoculated samples only).

Other vapor-phase treatments

Using a 2nd treatment apparatus, hydrogen peroxide vapor was applied by vaporizing 20 mL 35% H₂O₂ at 150 °C in a pressurized vessel and transferring the vapor to a 170-L vacuum chamber at 686 mm Hg vacuum in which sets of 4 inoculated apples

were placed in a colander. This apparatus was at room temperature and not in a water bath. Multiple cycles of this treatment were applied by adding successive charges of 35% H₂O₂ to the vapor generating vessel. Treated samples and uninoculated controls were handled as described above.

Chlorine dioxide vapor treatments were applied by direct exposure of inoculated apples to ClO₂ gas generated in sachets containing the proprietary precursor materials. The sachets were rated as to ClO₂ output over specified time intervals, based on analytical data obtained by ICA TriNova. Generation of ClO₂ gas was initiated by mixing compartmented portions of reactants within sachets immediately prior to each decontamination trial. Activated sachets were suspended from the underside of the lid of a 24.6-L (6.5-gal) high density polyethylene pail (M&M Industries, Chattanooga, Tenn., U.S.A.) containing a desiccator plate on which 4 apples were placed and to which 10 mL of water were added to maintain a constant humidity. Air circulation was provided by a battery-operated fan (Hankscraft Motors, Reedsburg, Wis., U.S.A.) attached to the underside of the lid. The screw-on lid was provided with a neoprene o-ring to give an air-tight seal. Chlorine dioxide gas concentrations in pails containing activated sachets were estimated using release profiles provided by ICA TriNova. Inoculated apples were exposed to ClO₂ doses between 0.75 and 7.5 mg, corresponding to maximum concentrations in the pails of 0.03 to 0.30 ppm, over exposure times between 1 and 20 h at 5 and 20 °C.

Microbiological methods

Treated apples and controls were weighed, cut into quarters on a sterile cutting board, combined with an equal volume (w/v) of sterile 0.1% peptone water (PW) (Difco, Detroit, Mich., U.S.A.), and homogenized for 1 min at medium speed in a sterile 4L stainless steel Waring blender (Waring Products Div., Dynamics Corp. of America, New Hartford, Conn., U.S.A.). Blended samples were filtered through a filter bag (Spiral Biotech, Bethesda, Md., U.S.A.) and the filtrate diluted as required with sterile PW. Generic *E. coli* counts were estimated by plating 0.1 mL aliquots on Trypticase Soy Agar (TSA) (Difco) with a spiral plater (Auto-plate 4000, Spiral Biotech). The TSA plates were overlaid after 2 to 3 h incubation at 35 °C with MacConkey agar (MAC) (Difco) to recover injured cells (Doyle and Schoeni 1984; Silk and Donnelly 1997).

Estimation of acetic acid and hydrogen peroxide uptake

Uninoculated treated apples and con-

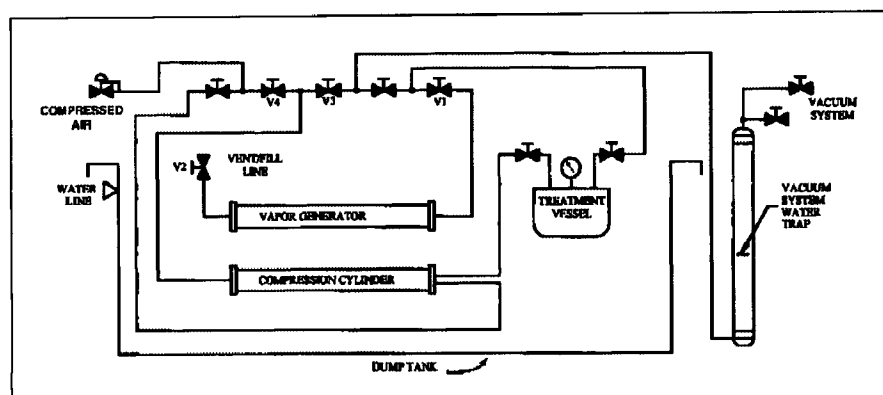


Figure 1—Acetic acid vapor generation and application system

trols (sets of 4) were juiced with an Acme Supreme Juicerator (Acme Supreme Juicer Mfg. Co., Waring Products Div., DCA, New Hartford, Conn., U.S.A.), lined with a Whatman nr 1 filter paper strip to exclude seeds, skin fragments, and coarse suspended solids from the juice. Aliquots of juice (50 mL), withdrawn by pipet to exclude foam, were mixed with 50 mL deionized water and titrated with 0.1 N NaOH to a pH 8 endpoint, measured with a Corning Model 130 pH meter. Acid uptake was calculated from the difference in titratable acid between treated samples and controls, expressed as acetic acid (% w/v).

Uptake of H₂O₂ per apple was determined following vapor treatment or immersion in 5% H₂O₂ by rinsing sets of treated, uninoculated apples with 2 L deionized H₂O and then estimating the concentration in the collected rinse water with the Reflectoquant Analysis System (EM Science, Gibbstown, N.J., U.S.A.).

Statistical analyses

Population reduction data were analyzed for differences in response to treatments by ANOVA, t-tests and the Bonferroni LSD test to separate means (Miller 1981). All statistical analyses were performed with SAS/STAT software (SAS Institute Inc 1989).

Results and Discussion

Modes of operation of vapor treatment apparatus with acetic acid

Using the results of the preliminary study at PARC (Delaquis and others 2000) as a point of departure, we applied acetic acid vapor treatments to inoculated apples to determine optimal modes of operation of the new vapor treatment system. Initial trials employed vacuum transfer of acetic acid vapor at 50 °C from the vapor generator to the sample treatment vessel over a 30-min exposure time. Population reductions in Golden Delicious apples inoculated with *E. coli* ATCC 25922 were between 2 and 3 logs (Table 1). As a 2nd treatment, this cycle was applied 4 times (about 7 min cycle time) over 30 min, but the results showed minimal improvement. As a 3rd treatment, the procedure was modified so that the transferred vapor was compressed to 103.4 kPa (15 psi) with the expectation that greater penetration of vapor into the fruit would occur. Application of repeated doses of vapor by means of successive vacuum/pressure cycles resulted in reductions exceeding 4 logs in 1 trial.

While significant reductions in *E. coli* population were obtained with these treatments, results from trial to trial were not con-

Table 1—Decontamination of Golden Delicious apples inoculated with *E. coli* (ATCC 25922) by pressurized acetic acid vapor

Treatment ^a	<i>E. coli</i> population reduction (log ₁₀ CFU/g) ^b		
	Trial 1	Trial 2	Mean
1 vacuum cycle	2.67 ± 0.00	1.99 ± 0.08	2.33 ^j
4 vacuum cycles ^c	2.99 ± 0.06	1.93 ± 0.08	2.46 ^{ei}
4 vacuum/pressure cycles ^d	4.19 ± 0.16	2.79 ± 0.04	3.49 ^e

^aAcetic acid vapor treatment conditions: vacuum = 686 mm Hg; pressure = 103.4 kPa temperature = 50 °C; total treatment time = 30 min. Each cycle includes evacuation of treatment vessel, transfer of vapor from vapor generator, and equilibration of treatment vessel at reduced pressure or compression to higher pressure for duration of cycle.

^bMean ± standard deviation; based on control population of 5.56 ± 0.06 and 4.95 ± 0.09 log₁₀CFU/g for Trials 1 and 2, respectively; enumerated on TSA/MAC.

^cTreatment vessel vacuum-filled 4 times with acetic acid vapor and held without compression for cycle time of about 7 min.

^dTreatment vessel vacuum-filled with acetic acid vapor; then pressurized to 103.4 kPa for cycle time of about 7 min; sequence repeated for total of 4 cycles over 30 min.

^{e-i}Means with no letters in common are different ($p < 0.05$) by the Bonferroni LSD mean separation test.

Table 2—Acid uptake by Golden Delicious apples treated with pressurized acetic acid vapor

Treatment ^a (vacuum/pressure)	Nr cycles	Time ^b (min)	Temperature °C	Rinse ^c	Acid uptake by juice ^d
686 mm Hg/103.4 kPa	4	30	50	None	0.588 ^e
686 mm Hg/103.4 kPa	4	30	50	Dip	0.526 ^e
686 mm Hg/103.4 kPa	4	7 to 10	50	Dip	0.104 ^f
686 mm Hg/103.4 kPa	1	5	50	Dip	0.58 ^f
508 mm Hg/34.5 kPa	1	5	60	Spray	0.058 ^f
508 mm Hg/68.9 kPa	1	5	60	Spray	0.048 ^f

^aTreatment applied by evacuating treatment vessel, breaking vacuum with hot acetic acid vapor, and pressurizing to specified pressure in single or multiple cycles.

^bTotal exposure time

^cDip in 6 L H₂O or spray with H₂O in colander

^dExpressed as % acetic acid (w/v), determined by subtraction of control titratable acidity from total titratable acidity; mean of duplicate trials

^{e-f}Means with no letter in common are different ($p < 0.05$) by the Bonferroni LSD mean separation test.

sistent. Furthermore, the apples showed some browning, and the juice had an atypical aroma. We suspected that the variability in treatment response seen in Table 2 and severity of treatment-induced defects were related to extent of acetic acid uptake by the apples. Estimates of acetic acid uptake, based on titration of juice expressed from treated and control fruit, clearly showed that the more prolonged treatments resulted in greater acid uptake (Table 2). Acid uptakes for 1 cycle (5 min) were 1/10 of those at 4 cycles (30 min), even at similar vacuum and pressure levels.

Discoloration of treated apples also was attributed to heat damage due to direct contact with the hot surface of the treatment vessel. Such contact and the resultant injury were eliminated by placing the apples in a colander within the treatment vessel.

Optimization of acetic acid vapor treatments

Treatment conditions were modified to avoid defects associated with excessive acetic acid uptake and injury from prolonged exposure or overheating by placing apples in a colander within the treatment chamber

and by reducing the vacuum to 508 mm Hg and the treatment time to 5 min, and log reductions for inoculated Golden Delicious samples were determined (Table 3). When a total treatment time of 5 min was used, log reductions increased as the treatment temperature and number of vacuum/pressure cycles were increased. Treatment pressure did not appear to affect the log reduction. Population reduction values exceeding 3 logs were obtained with 3 vacuum/pressure cycles at 60 °C at all pressures studied, but variability between replicate trials was high, indicating a need to identify and minimize, if possible, sources of treatment variability. Completely submerging the treatment vessel in the dump tank water appeared to reduce variability, both in acetic acid deposition and in treatment response (data not shown).

Slight darkening of lenticels was observed with Golden Delicious and Granny Smith but not with Fuji and Gala apples immediately after treatment at ≥ 60 °C. However, during storage for several hours, the Golden Delicious apples developed dark lesions surrounding the lenticels and penetrating several mm into the flesh beneath

the skin (Figure 2). Because of the severity of this defect, no further work was done on the acetic acid vapor treatments.

Hydrogen peroxide vapor treatments

When H_2O_2 vapor treatments were carried out over a range of operating conditions (incorporating variable dwell times, temper-

ature, vacuum and pressure levels, number of cycles) similar to those used with acetic acid, population reductions in inoculated apples were between 1.2 and 1.6 logs (data not shown), substantially less than would be obtained with a 5% H_2O_2 wash (Sapers and others 2002). Estimates of H_2O_2 deposition on treated apples, obtained by analysis of rinse H_2O , showed that exposure of apple

surfaces (and the attached bacteria) to H_2O_2 was much less with the vapor treatment than with a wash (Table 4). H_2O_2 vapor treatments applied with this system did not induce discoloration in Golden Delicious apples. We attempted to improve the efficacy of H_2O_2 vapor treatments by generating the vapor at 150 °C in a pressurized vessel and then transferring the vapor to a chamber, evacuated to 686 mm Hg, which contained the apples in a colander. Deposition of H_2O_2 increased to 34.3 mg/apple, more than resulted from washing, but population reductions increased only to 1.7 logs when 3 cycles were applied over 5 min. It likely that the high boiling H_2O_2 vapor condensed on the cool apple surface without contacting *E. coli* attached in the calyx area. No further

Table 3—Optimization of acetic acid vapor decontamination of Golden Delicious apples inoculated with *E. coli* (ATCC 25922)

Temperature (°C)	Nr cycles ^a	<i>E. coli</i> population reduction (log ₁₀ CFU/g) ^b		
		Pressure (kPa)		
		34.5	68.9	103.4
40	1	1.40 ^d	1.89 ^e	1.51 ^d
	3	1.93 ^d	2.30 ^{de}	2.10 ^d
50	1	2.08 ^d	1.82 ^e	1.50 ^d
	3	2.14 ^d	2.88 ^d	1.84 ^d
60	1	1.80 ^d	2.42 ^{de}	3.05 ^c
	3	3.52 ^c	3.70 ^c	3.47 ^c

^aTreatment applied by evacuating treatment vessel to 508 mm Hg vacuum, breaking vacuum with hot acetic acid vapor, and pressurizing to specified pressure in 1 or 3 cycles over total exposure time of 5 min.

^bMeans of duplicate trials; based on control population of 4.90 log₁₀CFU/g; enumerated on TSA/MAC

^{c-e}Within the same column, means with no letter in common are different ($p < 0.05$) by the Bonferroni LSD mean separation test.

Table 4—Deposition of H_2O_2 on surface of Golden Delicious apples by vapor-phase application or wash

Treatment	Nr of cycles	H_2O_2 deposition (mg/apple) ^a
H_2O_2 vapor- 508 mm Hg/68.9 kPa ^b	1	2.4 ^e
	3	4.0 ^e
H_2O_2 vapor- 686 mm Hg/68.9 kPa ^b	3	10.3 ^e
H_2O_2 vapor- 686 mm Hg at 150 °C ^c	3	34.3 ^d
5% H_2O_2 wash at 60 °C for 2 min	—	15.8 ^{de}

^aBased on recovery of H_2O_2 from duplicate sets of 4 treated apples where mean weight of the apples was 166 g.

^b H_2O_2 vapor treatment applied for 10 min at 60 °C.

^cVapor, generated by heating 20 mL 35% H_2O_2 to 150 °C in pressurized vessel, was transferred to evacuated chamber containing apples at 686 mm Hg; treatment repeated 2 more times for total exposure of 5 min.

^{d-e}Means with no letters in common are different ($p < 0.05$) by the Bonferroni LSD mean separation test.

Table 5—Decontamination of Golden Delicious apples inoculated with *E. coli* (ATCC 25922) by chlorine dioxide gas

Load size ^a	Treatment			Population reduction (log ₁₀ CFU/g)	Lenticel darkening
	Dose (mg ClO_2) ^b	Time (h)	Temperature (°C)		
4	7.5	3	20	3.24 ^d ^{ef}	Slight
	7.5	3	4	4.42 ^{de}	Slight
	7.5	20	20	4.91 ^d	Moderate
	7.5	20	4	5.00 ^d	Moderate
12	7.5	3	4	1.86 ^f	Slight
	7.5	20	4	2.65 ^{ef}	Slight
	15	20	4	3.85 ^{def}	Slight
	30	20	4	3.86 ^{def}	Slight

^aNr of apples in ClO_2 treatment pail

^bQuantity of ClO_2 released from sachet in treatment pail

^cMeans of duplicate or triplicate trials; based on control population of 5.36 ± 0.33 log₁₀CFU/g;

enumerated on TSA/MAC

^{d-f}Means with no letters in common are different ($p < 0.05$) by the Bonferroni LSD mean separation test.

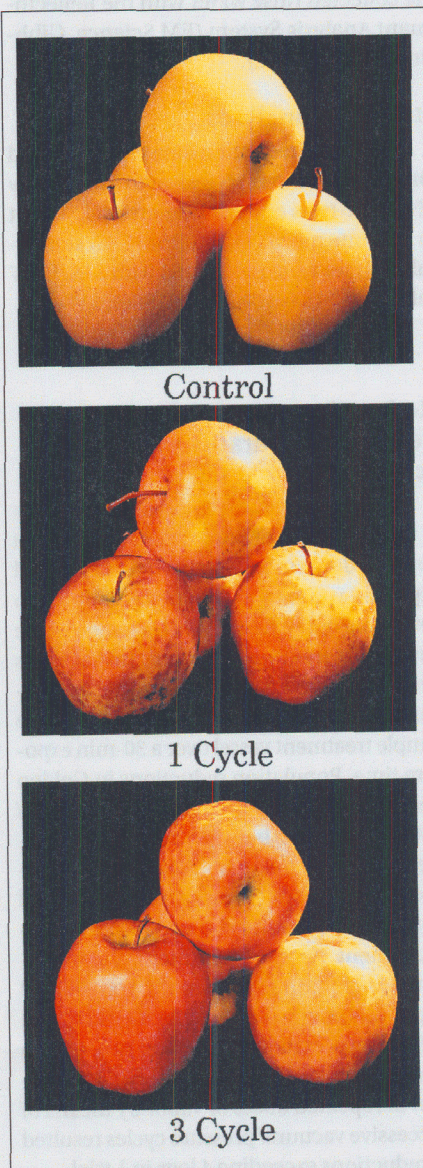


Figure 2—Golden Delicious apples treated with acetic acid vapor using 1 or 3 vacuum/pressure cycles of 508 mm Hg/68.9 kPa at 60 °C

work on treatment of apples with H_2O_2 vapor was carried out in this study.

Chlorine dioxide treatments

Application of ClO_2 gas to inoculated Golden Delicious apples generated with the vapor treatment apparatus operated at 60 °C with 3 cycles (508 mm Hg vacuum/68.9 kPa pressure) in 5 min resulted in a population reduction of $2.23 \log_{10}$ CFU/g ± 0.08 for 3 successive independent trials with no indication of injury to the fruit (data not shown). This result is comparable to population reductions obtained by washing with 5% hydrogen peroxide at 60 °C or 200 mg/L Cl_2 (Sapers and others 2000, 2002).

Exposure of inoculated apples to ClO_2 gas generated from sachets in an airtight pail was more effective in reducing *E. coli* populations than treatments applied with the vapor treatment apparatus. Population reductions were minimal (≤ 1 log) when a dose of 0.75 mg ClO_2 was applied (data not shown). However, when a dose of 7.5 mg ClO_2 was applied, the population reduction approached 5 logs (Table 5). Prolonged exposure to ClO_2 (20 h) resulted in darkening of lenticels, but this discoloration was minimal for 4 h exposures at 4 or 20 °C. A load effect was seen with ClO_2 gas treatment of apples, necessitating use of greater doses with increasing quantities of fruit to achieve a large population reduction. Research on optimization of ClO_2 gas treatments for apples and extension of this technology to other commodities is continuing.

Conclusions

ANTIMICROBIAL VAPOR TREATMENTS WERE investigated as a means of achieving greater inactivation of bacteria attached to apple surfaces due to improved contact with bacterial attachment sites. While acetic acid vapor treatments were capable of reducing *E. coli* populations on inoculated apples by as much as 3.5 logs, a large improvement over reductions achieved by sanitizer washes, the treatment must be considered infeasible because of severe discoloration of the treated fruit.

Hydrogen peroxide vapor has been used experimentally to produce decontamina-

tion. However, our results indicate that washing with dilute H_2O_2 solutions is considerably more effective than the vapor treatment, largely due to the relatively low volatility of H_2O_2 .

In contrast, our results with ClO_2 gas phase treatments indicated that a simple fumigation treatment, applied to fresh apples prior to packing for fresh market or fresh-cut processing, could achieve population reductions approaching FDA's mandated 5-log reduction for cider (FDA 2001). This is a 100-fold improvement over reductions obtained by washing with conventional sanitizers.

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